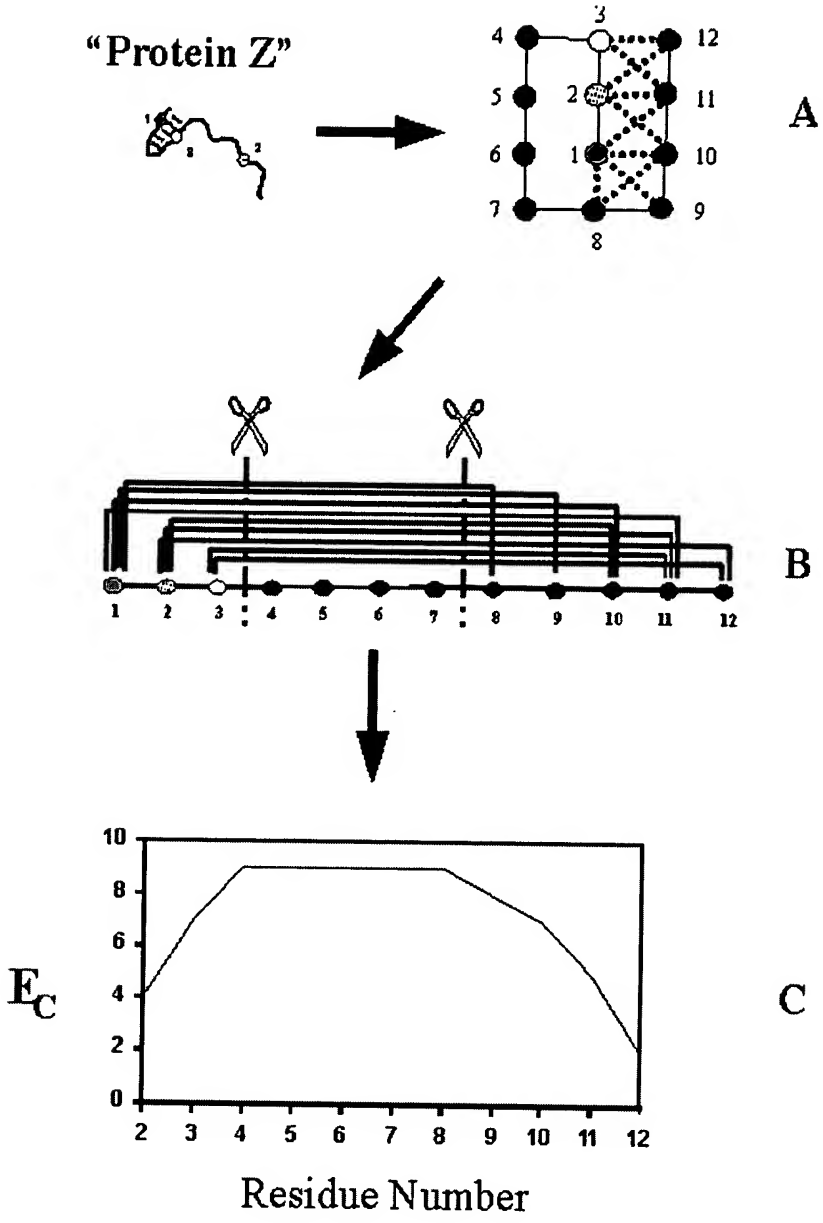
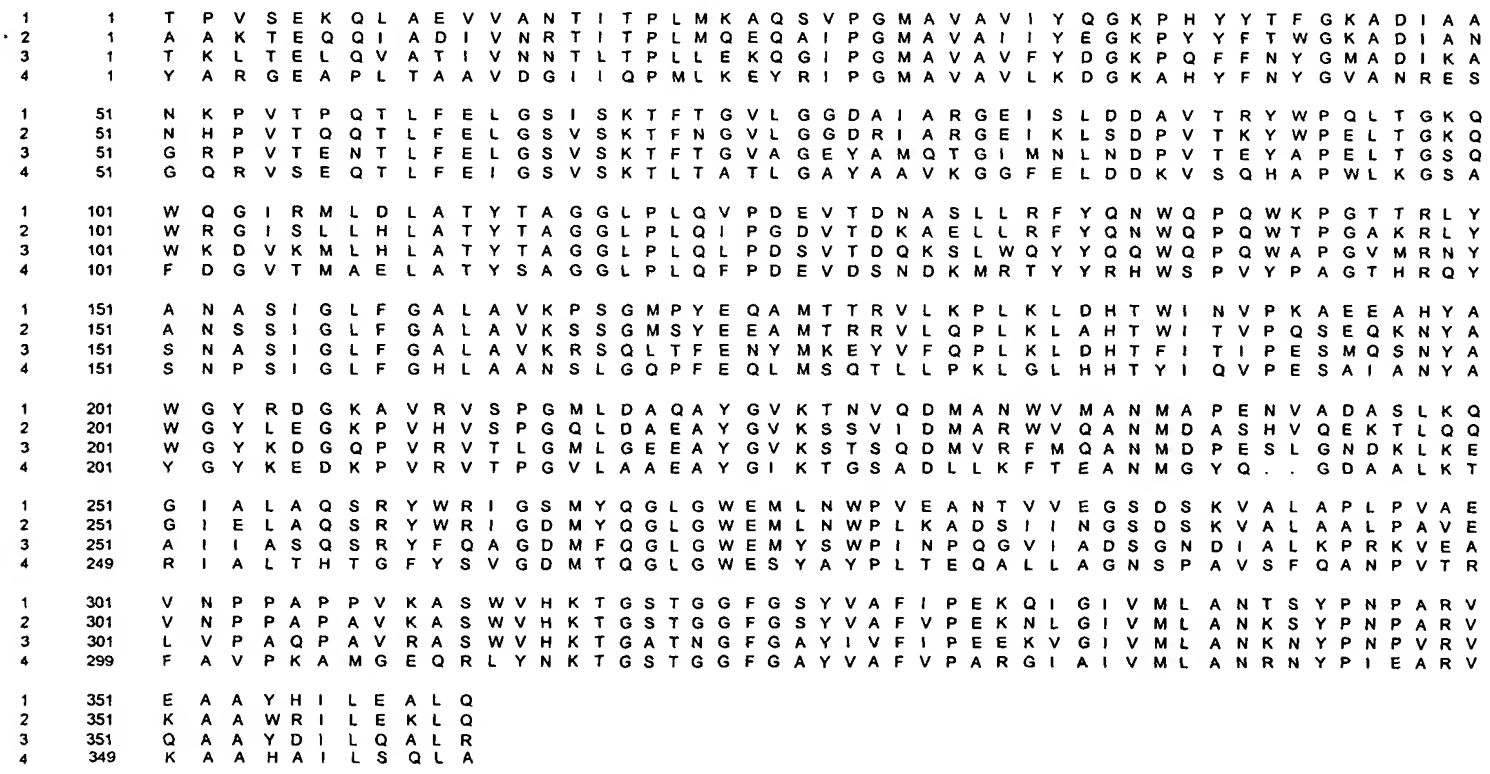
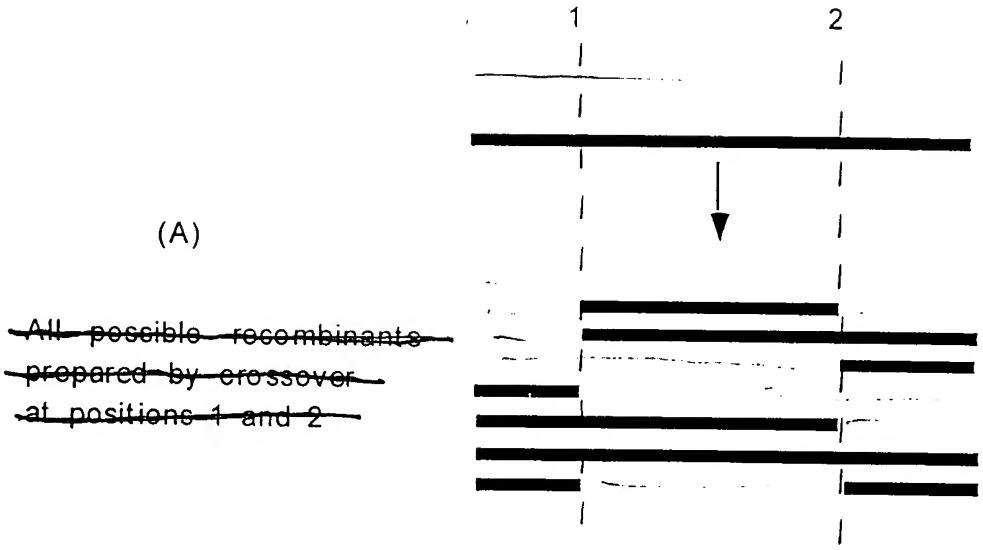


FIG. 2



1 Enterbacter cloacae P05364 ~~(X03866)~~ (SEQ ID NO:1)
2 Citrobacter freundii P05193 ~~(X07274)~~ (SEQ ID NO:2)
3 Yersinia enterocolitica P45460 ~~(X63149)~~ (SEQ ID NO:3)
4 Klebsiella pneumoniae Q48437 ~~(X77455)~~ (SEQ ID NO:4)

FIG. 3


1
2
(A)
All possible recombinants prepared by crossover at positions 1 and 2

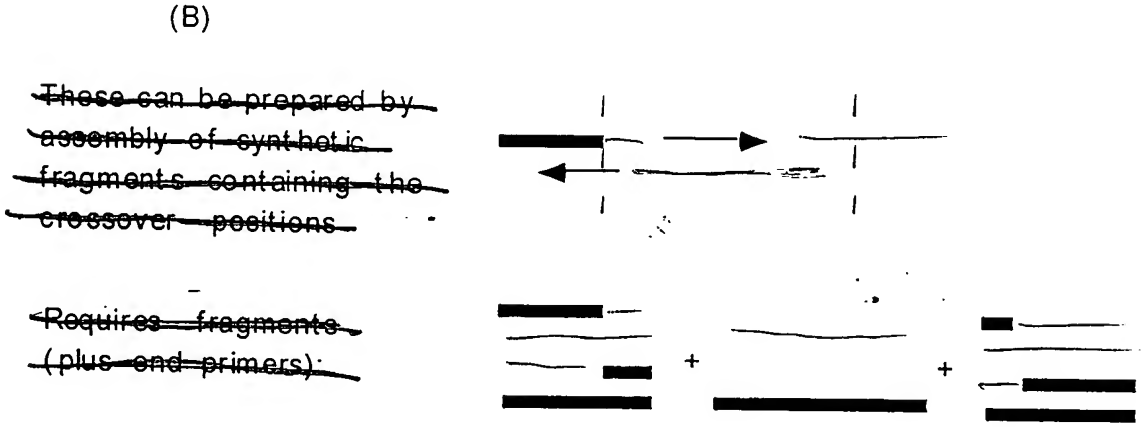

(B)
These can be prepared by assembly of synthetic fragments containing the crossover positions
Requires fragments (plus end primers)
+
+

FIG. 7

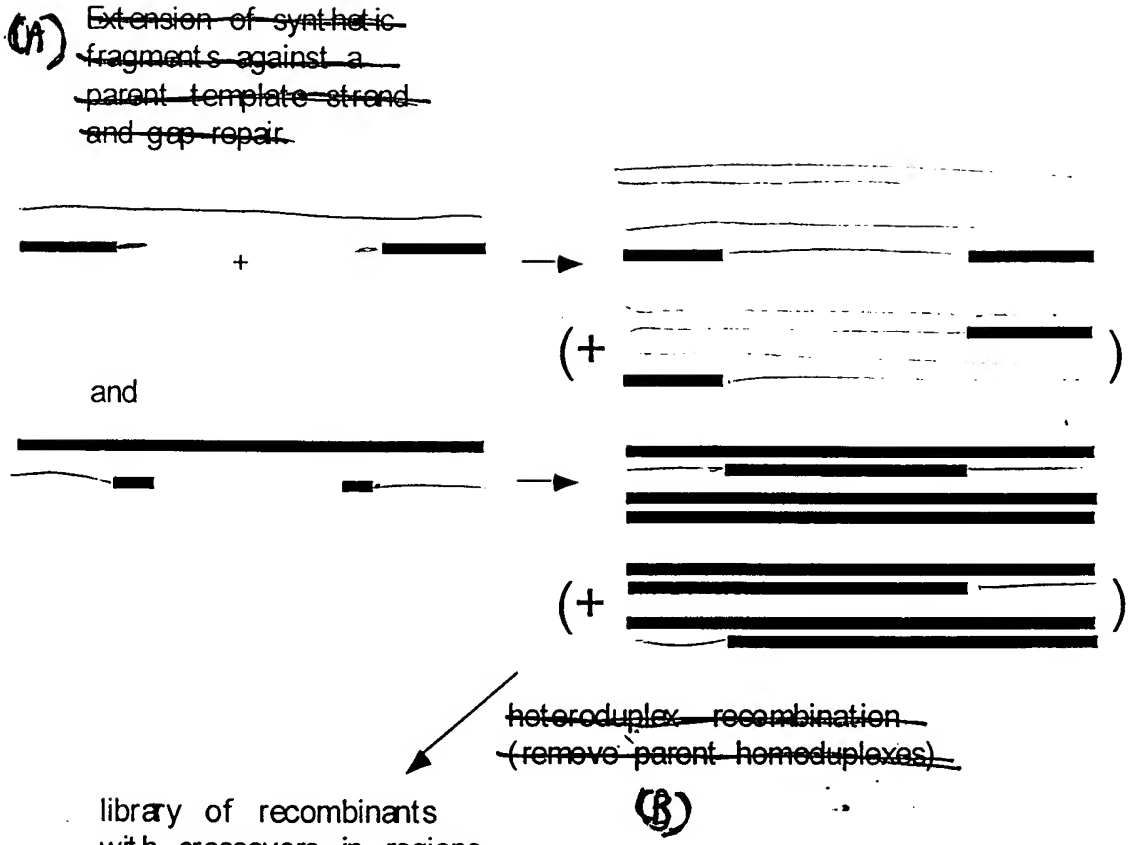
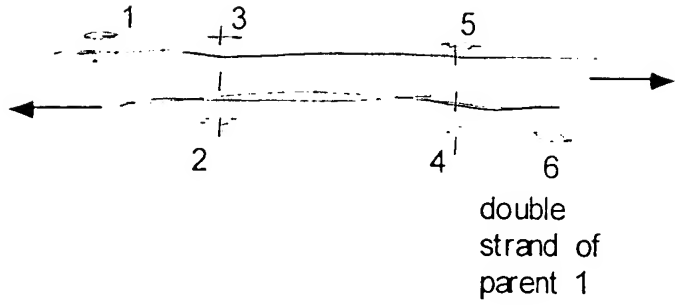


FIG. 8

(A)

~~Prepare the fragments by PCR with primers: perform reactions with primers 1+2, 3+4 and 5+6,~~

~~and do same for other parent(s).~~



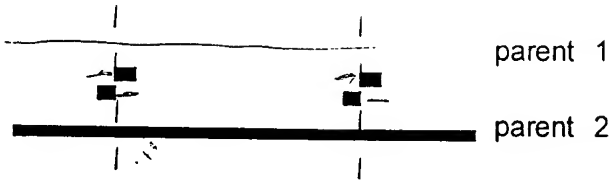
(B)

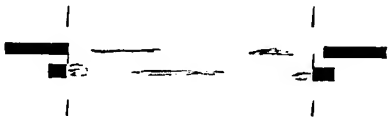
Reassemble fragments in a pool, by PCR with 1+ 6

FIG. 9

(A)

~~Prepare crossover primers designed to have crossovers at designated positions (2 primers for each position).~~





(B)

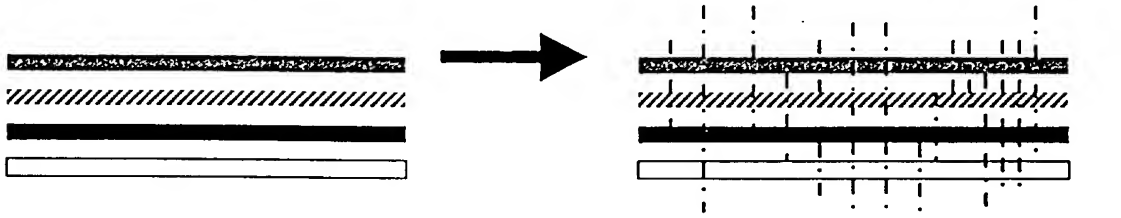
~~Fragment parent genes and PCR reassemble in the presence of the crossover primers to promote recombination at designated positions~~

FIG. 10

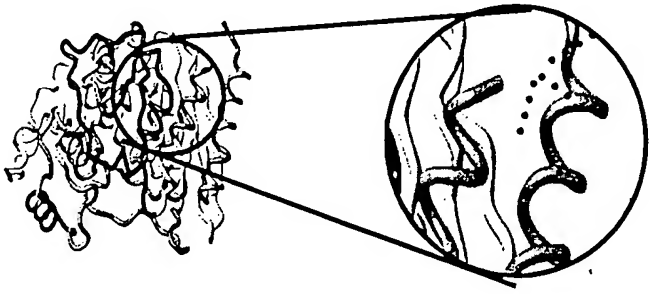
Recombinant search algorithm

1. Align parent sequences with template structure

2. Determine all possible crossover points according to sequence identity algorithm

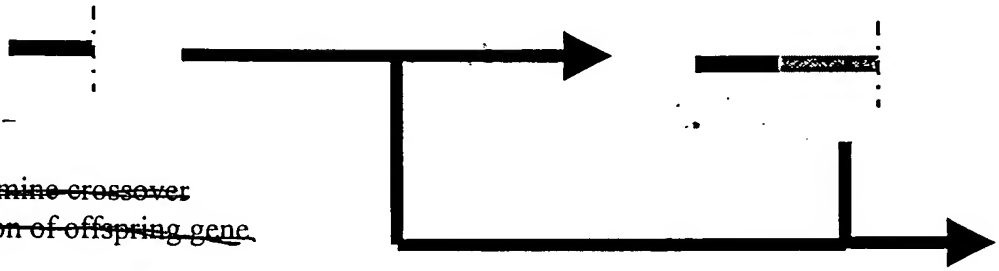


3. Calculate coupling matrix



4. Pick start parent at random and copy to offspring until a possible cut point is reached

5. Pick random number, if less than *p*, copy random new parent until next cut point is reached.



6. Determine crossover disruption of offspring gene.

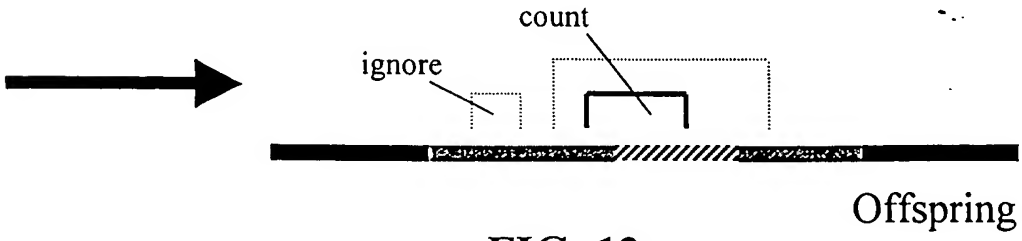


FIG. 12

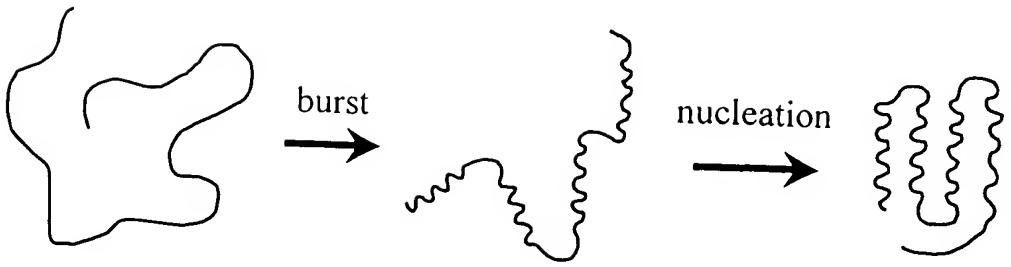


FIG. 18

~~The contact map shows residues that are distant (black) and residues that are close (white). If a given segment, [redacted], folds an above average number of residues into a given sphere size, then it is compact.~~

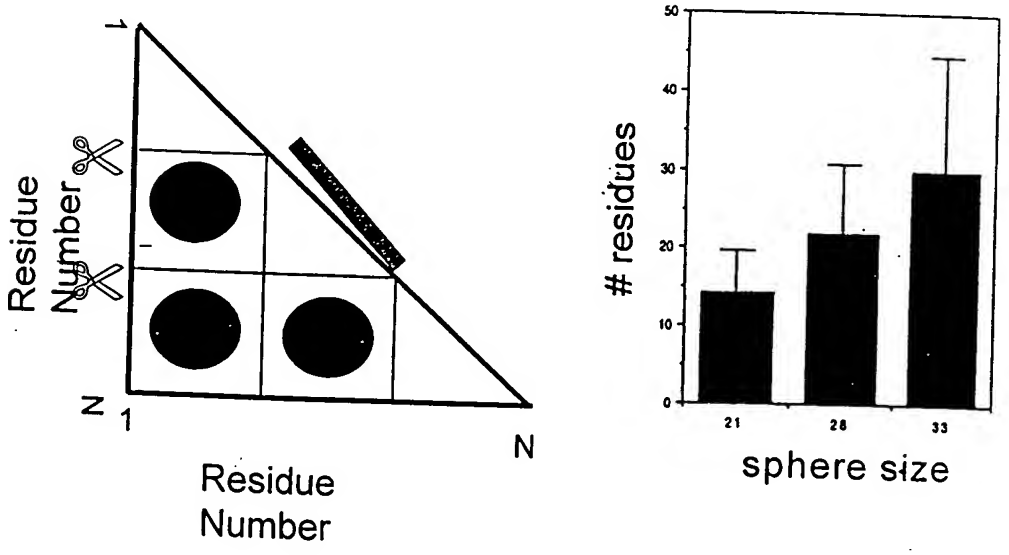


FIG. 19

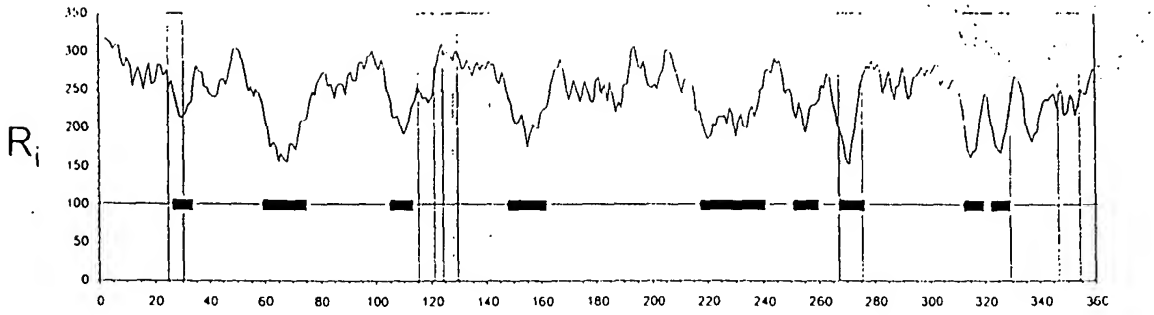
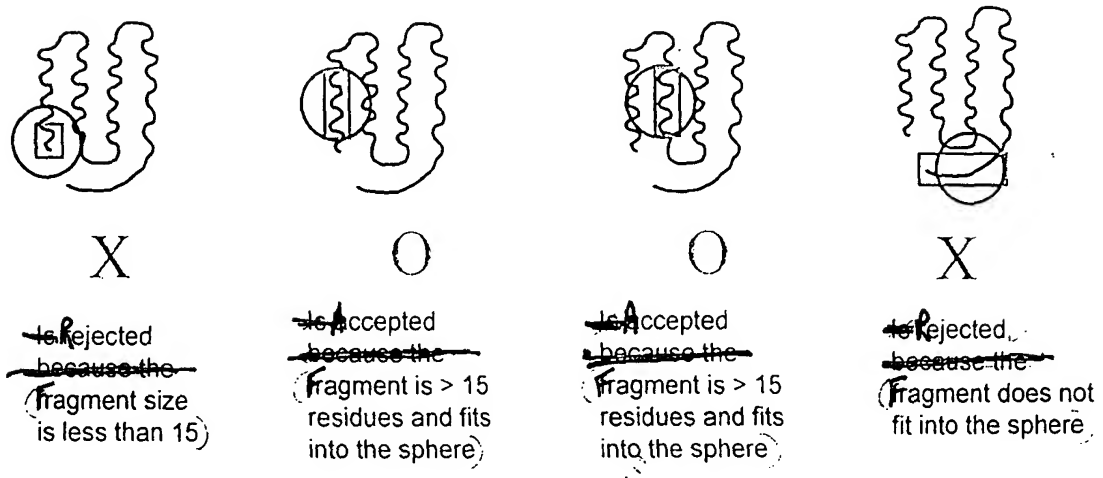


FIG. 22



(1) Pick a sphere size (21 angstroms, like Go-Gilbert) and a disruption threshold; (2) Scan protein using segments at least the average number of residues for that sphere size or greater (e.g., >15 for 21 angstrom sphere); 3) Check the disruption of all the compact fragments identified in step 2. If the fragment has a disruption above a threshold value, keep it; otherwise, throw it out; 4) If the compact unit is disruptive, increment the schema disruption measure for all of the residues in the fragment by one. This indicates that crossovers within the fragment are disfavored.

FIG. 23

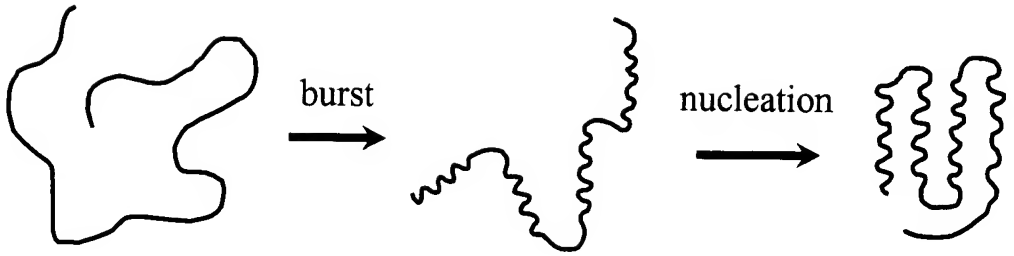


FIG. 18

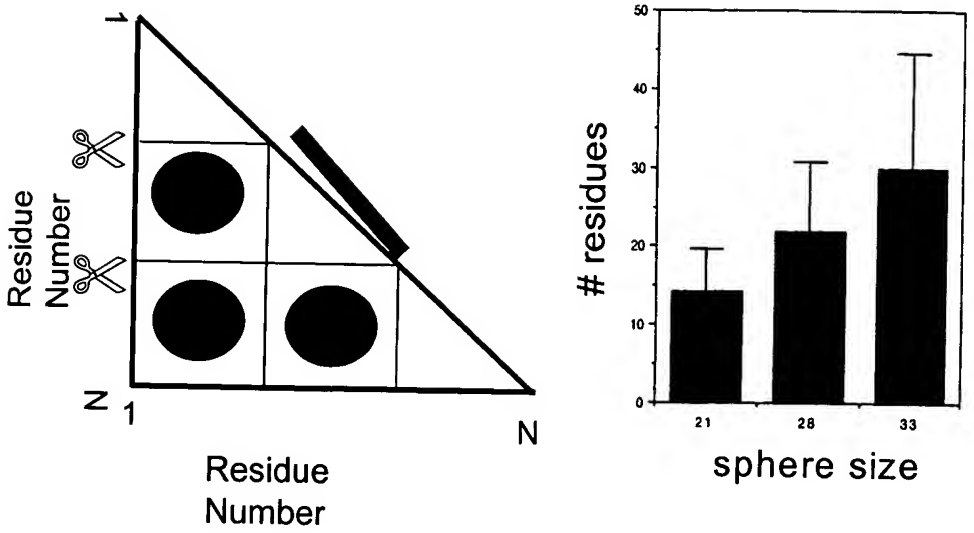


FIG. 19

Recombinant search algorithm

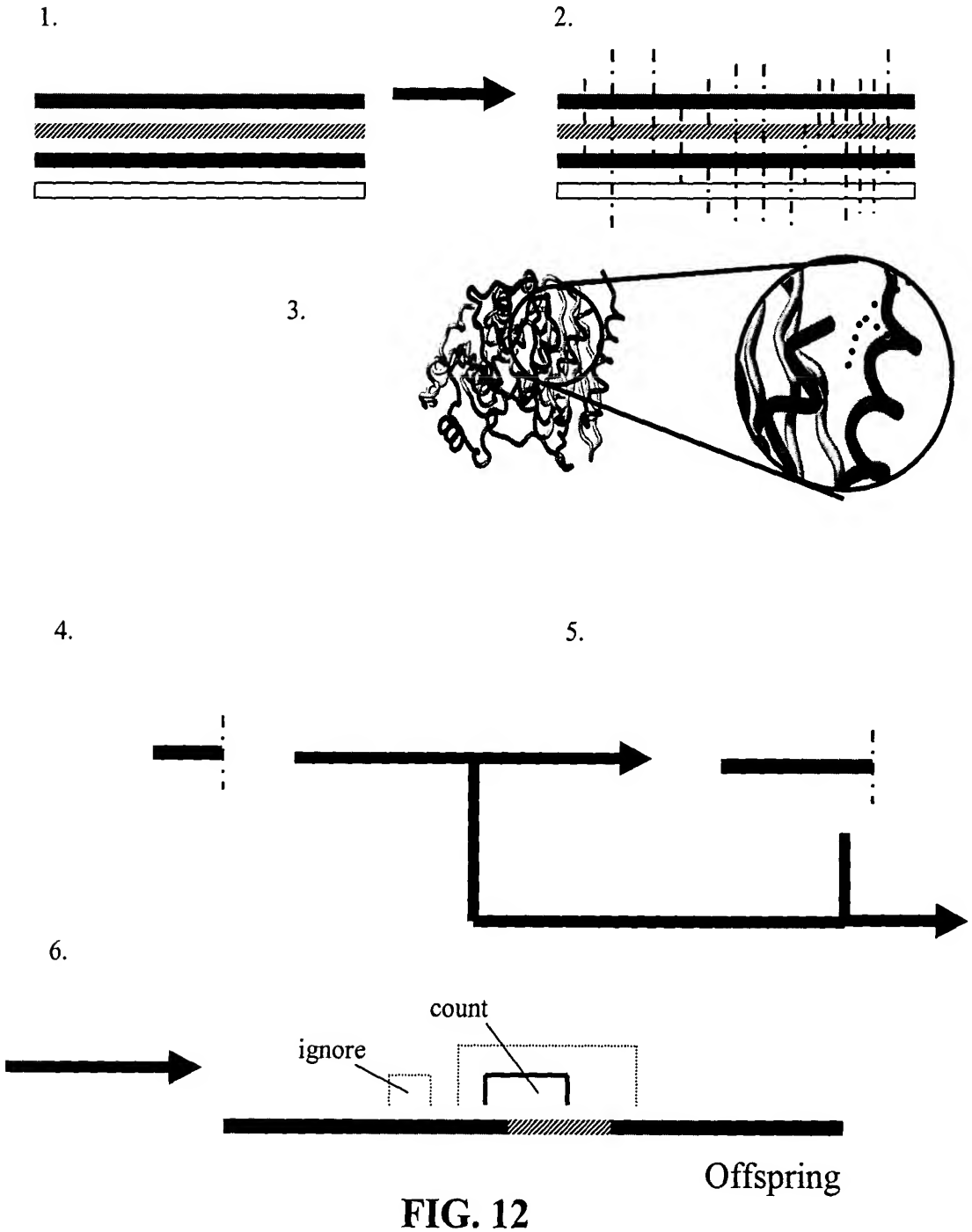


FIG. 12

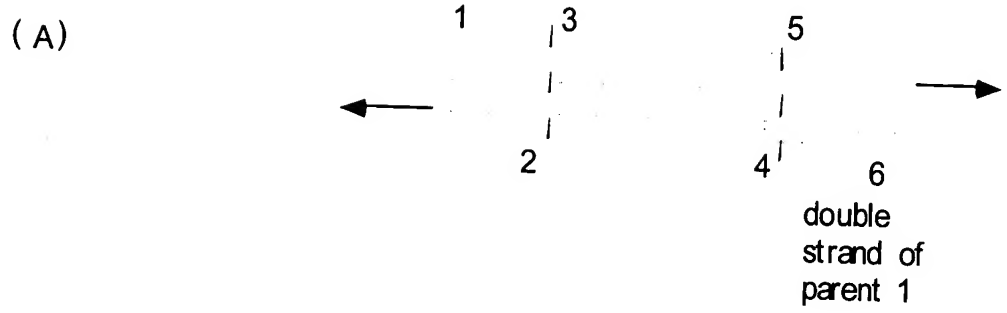


FIG. 9

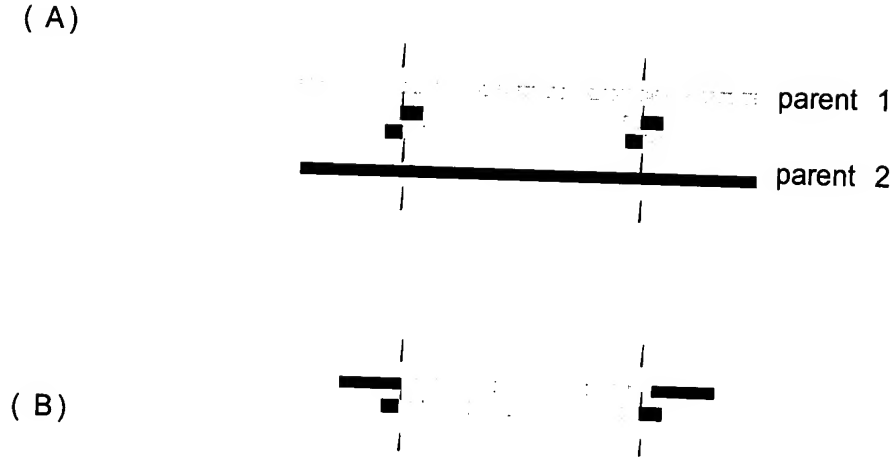


FIG. 10

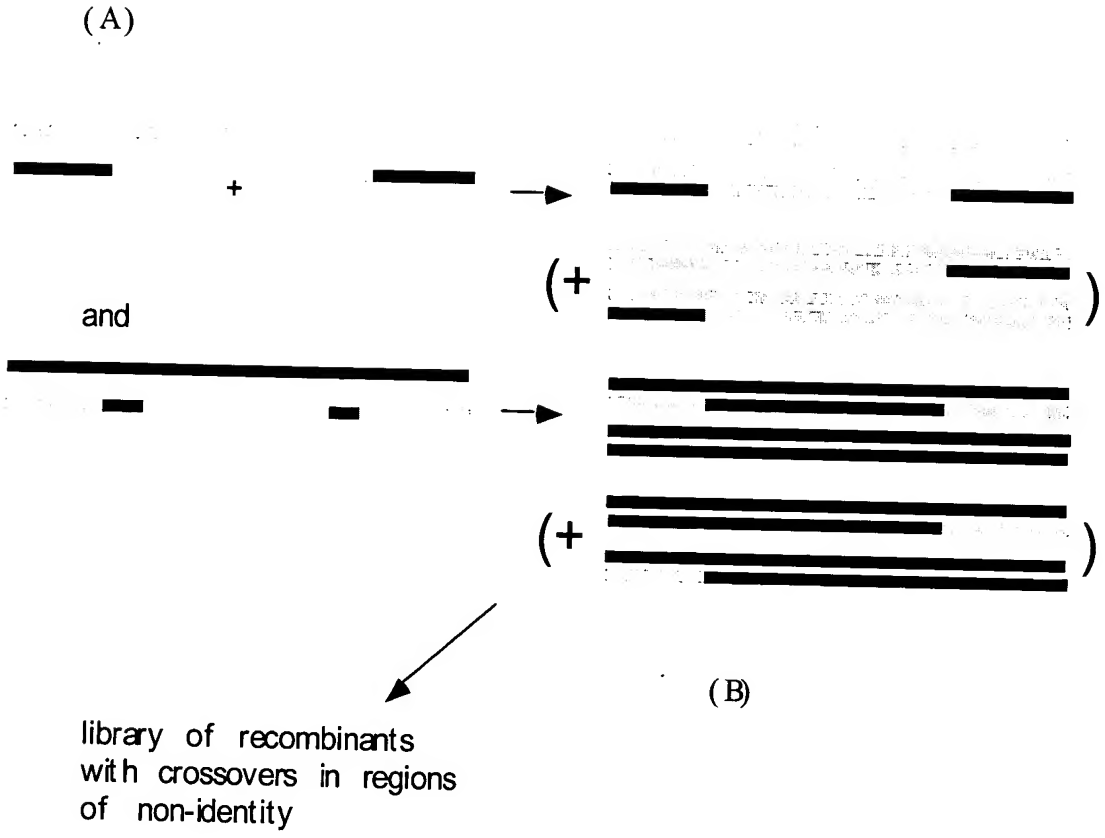
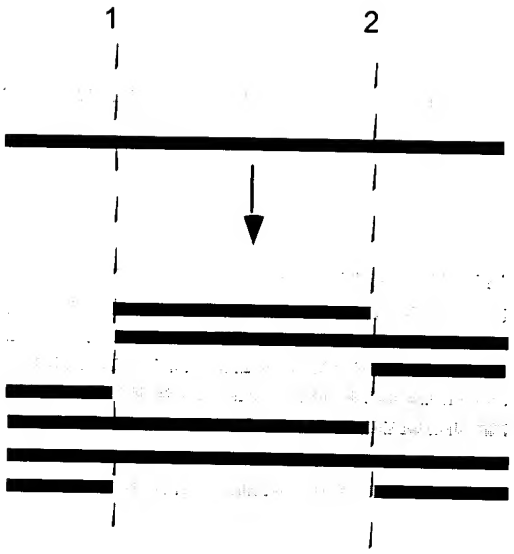


FIG. 8

(A)



(B)

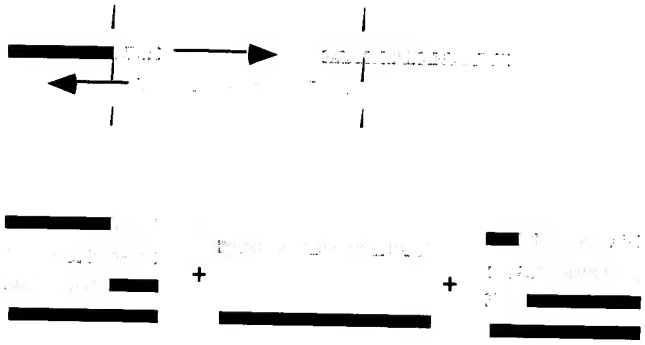


FIG. 7

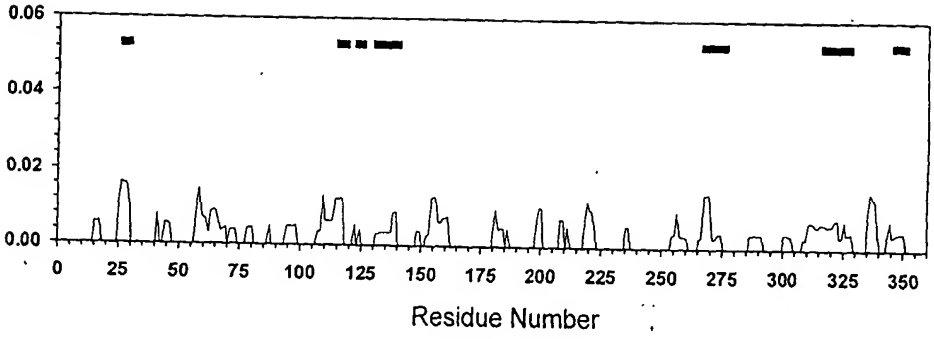

0.06
0.04
0.02
0.00
0 25 50 75 100 125 150 175 200 225 250 275 300 325 350
Residue Number

FIG. 4C

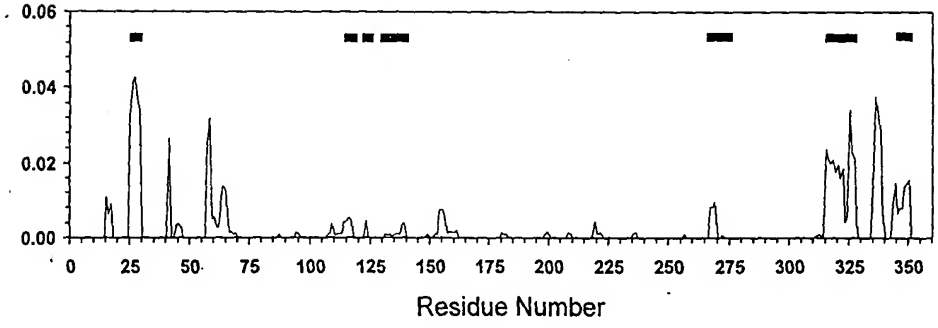
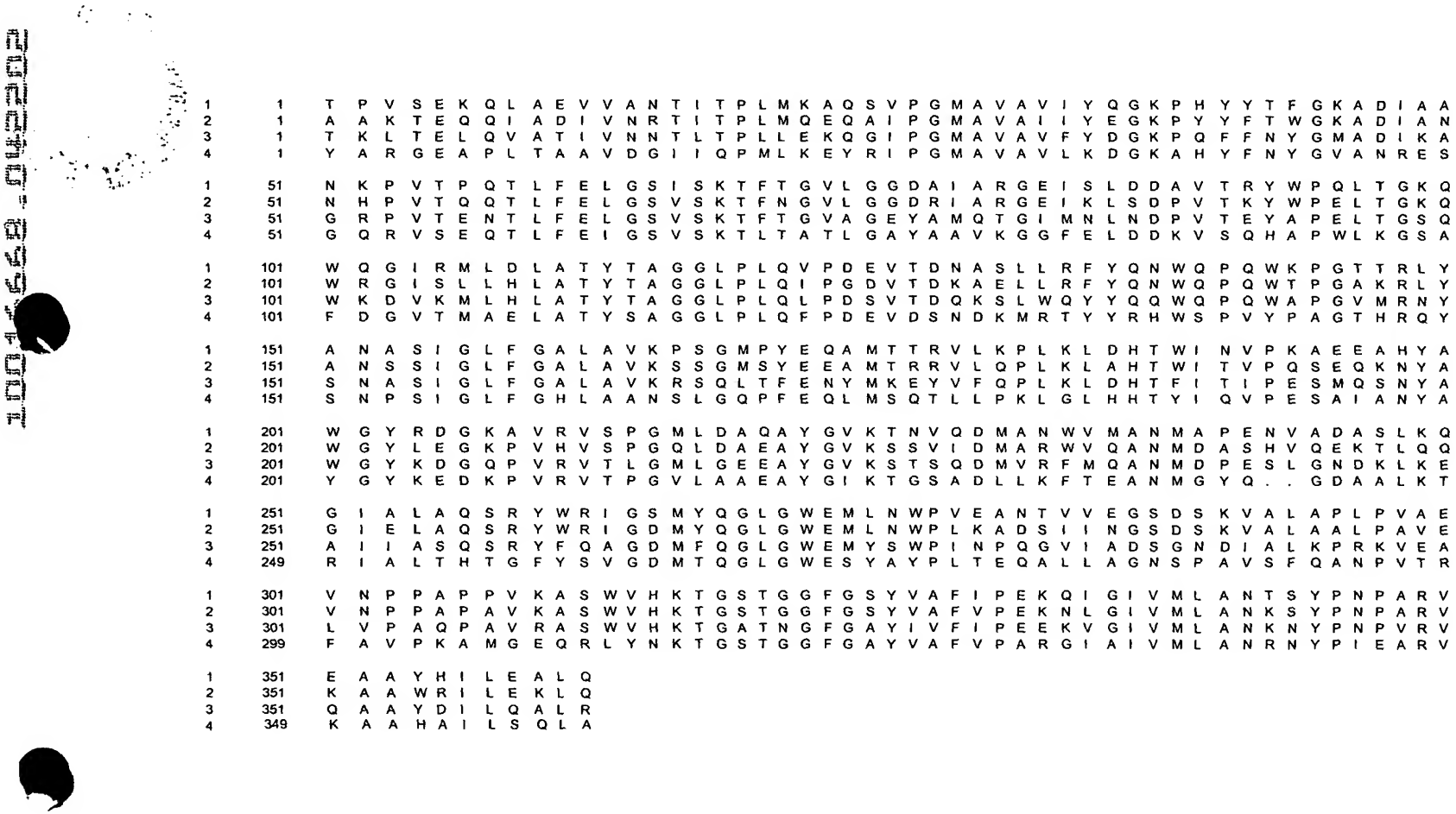

0.06
0.04
0.02
0.00
0 25 50 75 100 125 150 175 200 225 250 275 300 325 350
Residue Number

FIG. 4D



1 Enterbacter cloacae P05364 (SEQ ID NO: 1)
2 Citrobacter freundii P05193 (SEQ ID NO: 2)
3 Yersinia enterocolitica P45460 (SEQ ID NO: 3)
4 Klebsiella pneumoniae Q48437 (SEQ ID NO: 4)

FIG. 3

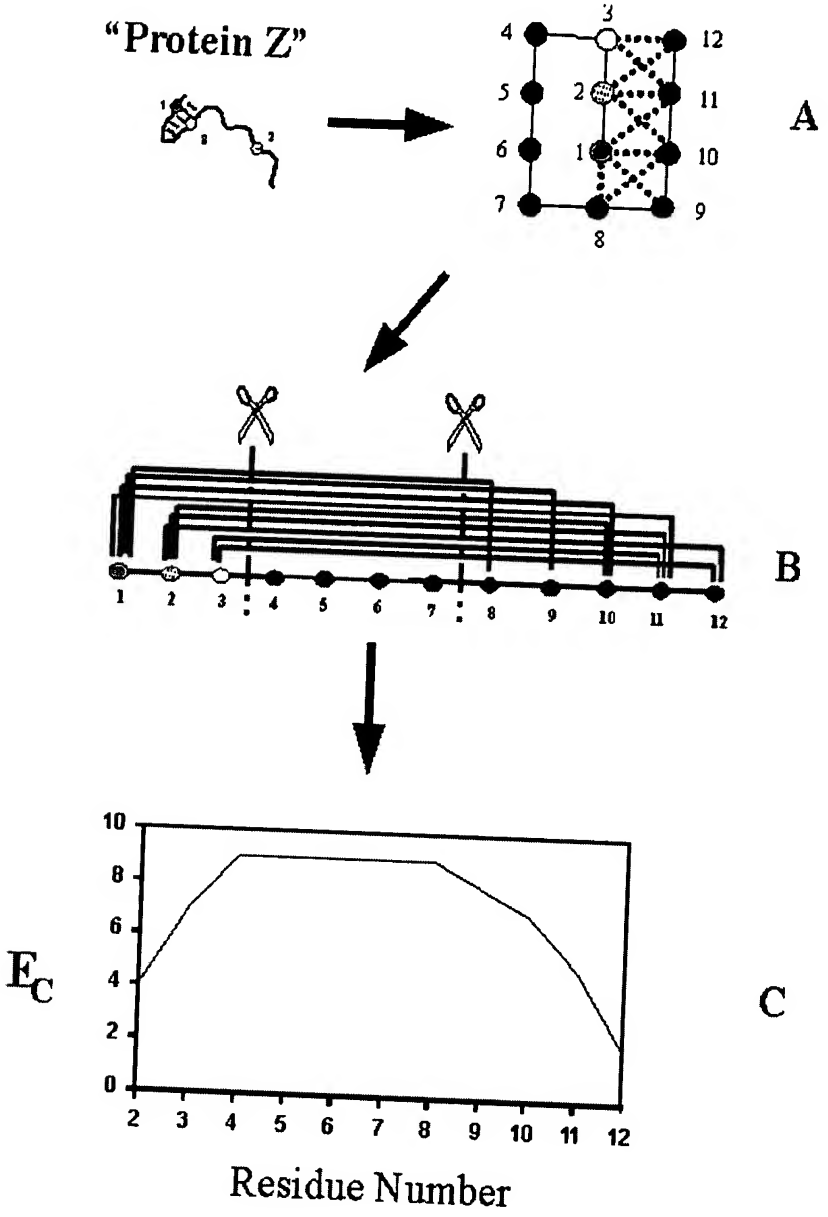


FIG. 2

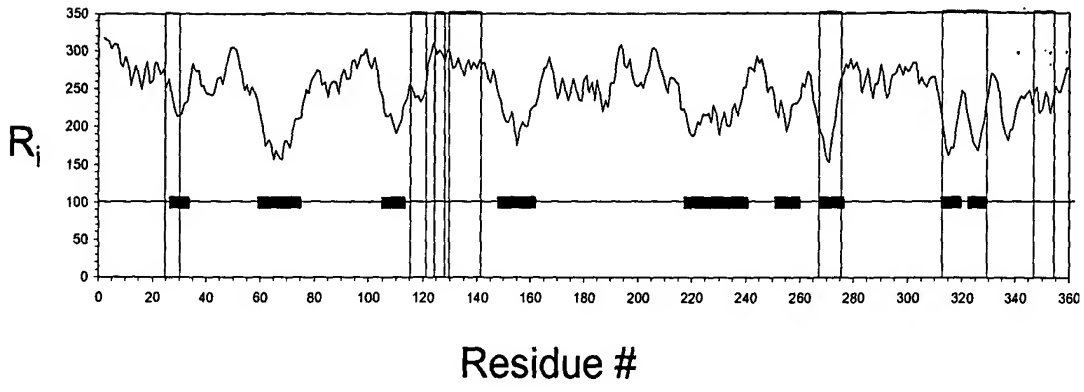

R_i
350
300
250
200
150
100
50
0
0 20 40 60 80 100 120 140 160 180 200 220 240 260 280 300 320 340 360
Residue #

FIG. 22

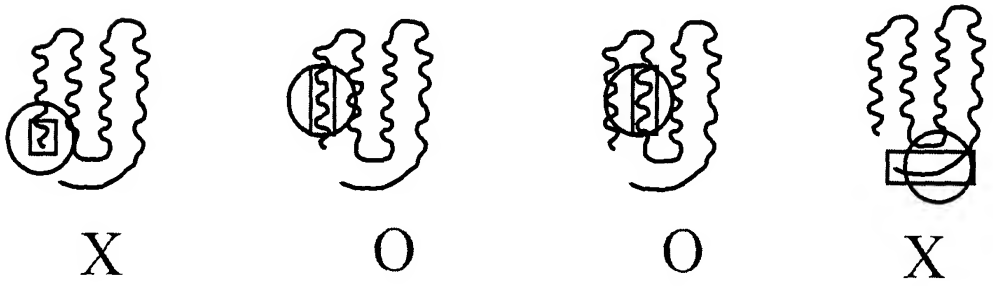

X
O
O
X
Rejected
Fragment size is less than 15
Accepted
Fragment is > 15 residues and fits into the sphere
Accepted
Fragment is > 15 residues and fits into the sphere
Rejected
Fragment does not fit into the sphere

FIG. 23